



Abstracts

Section IV Agriculture biotechnology

KN-012

Developing improved maize and soybean crops through transgenic and native traits

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Multiple approaches have been used to discover and develop traits to improve production agriculture crops. The technologies employed have included genetically based trait selections, such as screens of activation-tagged libraries of over-expressed genes, the testing of candidate gene leads developed through literature-generated hypotheses, through transcript and metabolite profiling experiments and/or through bioinformatic analyses, and native trait selection based on the incorporation of favorable alleles from diverse germplasm sources. For transgenic crop development, candidate genes are typically first tested in model plants. Gene shuffling technologies can be applied to screen for variation that can further improve trait functionality. Leads are transformed into elite germplasm and tested using surrogate screens in greenhouses. The transgenic plants are further assessed in managed and production field environments for reproducibility of the desired phenotype, for the absence of unwanted pleiotropic effects, and for yield potential. Alternatively, association mapping, linkage mapping and positional cloning technologies are employed to identify native genes that can be bred into elite lines, using molecular markers to introgress the trait of interest without unwanted surrounding genetic material; such products are non-transgenic. Products often combine transgenic and native traits. Robust intellectual property protection throughout the discovery and development processes is necessary to ensure that costs can later be recovered through trait premiums. To be competitive in the market, products need to carry multiple traits to confer tolerance to an ever-evolving array of pests and to abiotic stress factors, which has necessitated the development of trait stacking technologies. The use of molecular markers and doubled haploids in conjunction with contra-seasonal seed production accelerates trait integration and breeding timelines. For transgenic crops, field and laboratory data are assembled into complex regulatory dossiers for government approval processes. Finally, extensive field efficacy and multi-location breeding trials must be conducted to ensure that products meet performance standards.

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Gene expression, virulence and vaccine development in coronaviruses

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Coronavirus (CoV) transcription implies a discontinuous mechanism by which the 5'-terminal leader sequence is fused to the 5' end of the mRNA coding sequence (body). Transcription-regulating sequences (TRSs) preceding each gene include a conserved core, also found at the 3'-end of the leader, and variable 5' and 3' flanking sequences. Base pairing between the leader TRS (TRS-L) and the complement of the body TRS (cTRS-B) in the nascent RNA is a main determinant factor during CoV transcription. In transmissible gastroenteritis CoV, a good correlation has been observed between subgenomic mRNA levels (sgmRNA) and the free energy of TRS-L and cTRS-B duplex formation, with the only exception of sgmRNA N, the most abundant during viral infection in spite of its minimum free energy. Consequently, we postulated the presence of additional factors that regulate transcription of sgmRNA N. In fact, we have demonstrated the presence of a transcription enhancer preceding the coding sequences of N gene. These sequences have an enhancer activity not previously described within the *Nidovirales* order. SARS-CoV attenuated phenotypes were engineered in which the structural E gene (delta E), the group specific genes 6, 7a, 7b, 8a, 8b and 9b (delta 6–9), or E plus the group specific genes (delta E, 6–9) were deleted using an infectious cDNA clone. Viral particles with a morphology similar to that of the parental virus were observed in monkey cells in all cases. The virulence and induction of protection by the mutant viruses have been evaluated in two animal models: hamsters and transgenic mice expressing the SARS-CoV receptor hACE-2. The delta E virus was attenuated in hamsters and transgenic mice, and provided protection against homologous and heterologous SARS-CoV strains in both animal models. The data indicates that E gene is a virulence factor, and that viruses in which this gene has been deleted are promising vaccine candidates.

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GMO testing: Past, present and future perspectives

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A review of GMO testing methods from the first screening tests to prospective technologies with a potential to cope with tomorrow's analytical challenges (Hernandez et al., 2005; Holst-Jensen, 2007) is presented. Fitness for purpose is the number one issue when analytical methodology is chosen. A simple but very limited confirmatory test can be performed with a dipstick ELISA targeting a novel protein within minutes at low cost and with minimum requirements for equipment and training of personnel. A GMO identification and quantification test to determine if a sample contains any unauthorised GMO, or whether the GMO concentration exceeds a specified threshold, on the other hand can be very complex and resource demanding (Cankar et al., 2008; Holst-Jensen et al., 2006; Tengs et al., 2007). Stakeholders through the production chain have divergent priorities, but transparency and harmonised terminology is crucial to avoid conflict. Published literature on GMO testing is reviewed. Stakeholders and their needs are defined. A terminology to facilitate transparency and communication is proposed. The possibilities and limitations of different technologies are highlighted, bearing in mind the evolving and divergent stakeholder needs. Isolated focus on individual stakeholder interests is incompatible with transparency and communication among stakeholders in the production chain (a farm-to-fork perspective). Choosing between analytical methods is therefore complicated. Balancing interests of divergent stakeholders is controversial, but proper understanding of the possibilities and limitations of technologies may help decision makers.

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Brown midrib sorghum for second-generation ethanol production

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Sorghum [*Sorghum bicolor* (L.) Moench] stover can provide an abundant alternative source of fermentable sugars through enzymatic hydrolysis (Vermerris et al., 2007). While production of cellulosic ethanol from stover is feasible from an energy-balance perspective, its production is currently not economically viable. Improvements in bio-processing, technologies coupled with development of high biomass yielding genotypes with low lignin content make ethanol production cost effective. The brown midrib (*bmr*) mutant sorghum lines have significantly lower levels of lignin content (51% less in stems and 25% less in leaves (Porter et al., 1978). Therefore, the use of *bmr* cultivars would reduce the cost of biomass-based ethanol production by reducing pre-processing costs. ICRISAT has developed 11 female parents (A-/B- lines) and 22 pollen parents (R-lines) using *bmr* 1, *bmr* 3 and *bmr* 7 sources for development of high biomass *bmr* sorghum hybrids (Reddy et al., 2008). Preliminary evaluation of *bmr* hybrid parents (4 R-lines) derived from *bmr* 1 source (IS 21887) indicated 20% lower lignin content on whole plant basis compared to source (4.24%). Further, two brown midrib sources *bmr* 6 (reduced activity of cinnamyl alcohol dehydrogenase) and *bmr* 12 (reduced activity of caffeic acid O-methyl transferase) are under use in breeding program for *bmr* introgression. With several *bmr* mutant sources available in gene bank, ICRISAT has a comparative advantage to develop high biomass-yielding *bmr* sorghum hybrids for enhancing ethanol production from stover.

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Global adoption, impact and future prospects of biotech/GM crops

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In the early 1990s, some were skeptical that genetically modified (GM) or transgenic crops, now more often referred to as “biotech crops”, could deliver improved products and make an impact at the farm production level. There was even more